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TITLE: Reversing Maladaptive Plasticity to Cure Autonomic Dysreflexia after Spinal Cord Injury

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14. ABSTRACT Autonomic dysreflexia (AD) is a potential life threatening condition characterized as episodic vascular hypertension (often with bradycardia) that develops in most people with a spinal cord injury (SCI) above thoracic spinal level T5. Using telemetric recording we were able to detect biphasic spontaneous AD developed in mice with T3 SCI; the early phase of AD occurs within first week which is likely due to loss of descending control of sympathetic outflow and the late phase occurs weeks post injury which is likely caused by the formation of aberrant sympathetic neural circuits at the site of injury. We proposed that post-injury inhibition of reactive synaptogenesis would block the onset or reduce the severity of AD. In this study we tested this hypothesis by using both genetic modified mice lines ($\alpha 2\delta$ -1 KO, $\alpha 2\delta$ -1 over-expressing and TSP KO) and the drug, Gabapentin, which disrupting the binding of $\alpha 2\delta$ -1 with TSP, to block the formation of aberrant sympathetic nerve circuits and prevent occurring of AD. Current study suggested the $\alpha 2\delta$ -1 over-expressing mice show increased AD events in the early phase and the late phase of AD is not developed in 129 mice. However both TSP4 KO mice and WT littermates developed significant amount of AD start from first week after injury. In the future we are going to test effect of GBP on the development of AD.					
15. SUBJECT TERMS Spinal cord injury (SCI); Autonomic dysreflexia (AD); Synaptogenesis; Sympathetic pre-ganglionic neurons (SPNs); Gabapentin (GBP); Thrombospondins (TSP); Calcium channel subunit a1d2.					
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INTRODUCTION

Narrative that briefly (one paragraph) describes the subject, purpose and scope of the research.

Autonomic dysreflexia (AD) is a life threatening condition of episodic vascular hypertension (often with bradycardia, i.e., slowed heart rate) that develops in most (~90%) people with a spinal cord injury (SCI) above thoracic spinal level T5. After high level SCI, loss of supraspinal control together with aberrant collateral sprouting and formation of new intraspinal synapses (i.e., synaptogenesis) causes spinal autonomic reflexes to become exaggerated [1-4]. This post-injury maladaptive neural plasticity, involving sensory axons and propriospinal interneurons that connect multiple segments of the thoracic and upper lumbar spinal cord, progresses slowly over the course of several weeks or months post-injury. Prevention (e.g., regular bladder/bowel care) and anti-hypertensive medications are currently the best way to “manage” AD; however, there is no cure [5]. In this study, genetic and pharmacological tools are used to test the hypothesis that post-injury inhibition of reactive synaptogenesis will block the onset or reduce the severity of AD. After CNS injury, astroglia and macrophages secrete thrombospondins (TSP), a family of matricellular proteins that regulate cell-cell and cell-matrix interactions, most notably neurite growth and synaptogenesis [6, 7]. Eroglu et al. showed that astrocyte-derived TSPs cause synaptogenesis by binding to neuronal $\alpha 2\delta$ -1 receptors and that transgenic over expression of neuronal $\alpha 2\delta$ -1 dramatically increases synaptogenesis [7]. TSP-4 is selectively increased in astrocytes surrounding injured spinal cord axons. Here with genetic tools, we predict that either TSP-4 or $\alpha 2\delta$ -1 knockout will block the onset or reduce the severity of AD, however the $\alpha 2\delta$ -1 over-expression will enhance the development of AD. Anti-epileptic/anti-neuropathic pain drugs gabapentin (Neurontin) and pregabalin (Lyrica) bind with $\alpha 2\delta$ -1 [8] thereby blocks TSP/ $\alpha 2\delta$ -1 interactions as well as inhibits TSP-induced new synapse formation[7]. We hypothesize that inhibiting TSP binding to neuronal $\alpha 2\delta$ -1 with GBP will reduce the severity and frequency of AD by inhibiting maladaptive synaptogenesis after SCI. Current study suggested the $\alpha 2\delta$ -1 over-expressing mice show increased AD events in the early phase. Both spontaneous AD (sAD) and induced AD (IAD) are not developed at the late stage in 129 mice line, which is the genetic background of $\alpha 2\delta$ -1 mice. Both TSP4 KO and WT littermate developed significant numbers of Ad even at first week after T3crush, but there is no significant difference between WT and TSP KO. In the future we are going to test the effect of GBP on the development of AD post T3 SCI.

KEYWORDS: *Provide a brief list of keywords (limit to 20 words).*

Spinal cord injury (SCI), Autonomic dysreflexia (AD), Thrombospondin (TSP), $\alpha 2\delta$ -1 receptor, Gabapentin (GBP)

ACCOMPLISHMENTS

What were the major goals of the project?

- Goal 1: Comparing AD events in the three mice strains ($\alpha 2\delta$ -1 TG; $\alpha 2\delta$ -1 null; TSP null) vs their respective littermate after T3 SCI. (60% completion)
- Goal 2: Comparing AD events in GBP vs control group after T3 SCI. (30% completion)
- Goal 3: Complete immunohistochemistry analysis for synaptogenesis of all animals. (15% completion)
-

What was accomplished under these goals?

Goal 1: Comparing AD events in three mouse strains ($\alpha 2\delta$ -1 TG; $\alpha 2\delta$ -1 null; TSP null) vs their respective littermates after T3 SCI. (60% completion)

1) Major activities and Specific objectives:

Breeding/expanding mouse colonies: We obtained $\alpha 2\delta$ -1 TG and TSP null mice lines from Dr Eroglu (Duke) on Feb 7, 2014. After 8 weeks of quarantine, both mice lines started breeding. In August 2014, enough mice (n=10 for $\alpha 2\delta$ -1 TG and n=9 for WT littermates) were available to allow us to evaluate the development of AD (frequency and/or severity) in $\alpha 2\delta$ -1 TG vs WT littermates after T3 SCI. Initially, there were problems breeding TSP4 KO mice but by Jun 2015, 10 TSP-null mice and 6 control littermates were available to compare the development of AD in TSP KO vs WT littermates after T3 SCI.

Surgery procedures: All animals were maintained in a pathogen-free environment. Telemetry transmitters were implanted into each mouse as described previously [9]. Briefly, to monitor the blood pressure (BP) and heart rate (HR), the PhysioTel telemetry system with PA-C10 telemetry transmitters (Data Sciences International) was implanted in anesthetized mice 3 days before SCI via a cannulation of the left common carotid artery (CCA). The extra-vascular portion of the transmitter was placed into a subcutaneous pocket created on the lateral flank. The CCA was exposed through a midline incision on the neck, and the catheter of transmitter was introduced into the CCA through a small incision near the carotid bifurcation and advanced until the sensing region of the catheter was positioned in the aortic arch (8–9mm from the carotid bifurcation). Complete spinal cord transection injuries were performed as described previously [10]. Briefly, using aseptic technique, a partial laminectomy was performed at vertebral level T3, after which the periosteum and dura mater were carefully opened. Using iridectomy scissors together with gentle aspiration, the spinal cord was cut, creating a clear separation between the rostral and caudal stumps of transected spinal cord. After injury, muscle and skin were sutured separately and then mice were injected with sterile saline (2 ml, s.c.) and placed individually into warmed HEPA filtered cages. Postoperative care included manual bladder expression 2/d and daily antibiotics for the first 7day post injury (dpi) (gentocin, 5mg/kg, s.c.) and manual bladder expression 2X/day until euthanized. Dehydration was monitored daily and body weight and

urinary pH was monitored weekly. All surgical procedures were approved by and performed in accordance with the Institutional Laboratory Animal Care and Use Committee at The Ohio State University.

Data acquisition, processing and Analysis of spontaneous AD: The data from the TA11PA-C10 device were transmitted via radio frequency signals to a receiver below the home cage and collected using the Dataquest A.R.T. system, version 4.2 (Data Sciences International). The data were recorded continuously day and night with sampling rate of 500 Hz. MATLAB software was used to create an algorithm that would detect episodes of spontaneous AD. The current automated program works similarly to a previous semi-automated method developed in our lab [9] but incorporates manual validation steps into the computer algorithm to improve objectivity and efficiency (see Figure 1). Specific algorithm parameters that are screened to detect dysreflexic episodes include a blood pressure increase over baseline generated with lowess smoothing curve (scan 6 minute time window) of 10 mmHg (H1) and a heart rate decrease over baseline of 10 BPM (H2). H3 (20 mmHg) and H4 (30 BPM) represent minimum variations in blood pressure and heart rate based on diagonal line segments spanning the onset and offset of the blood pressure increase (T1) and heart decrease (T2). The determination of the onset and offset of these segments is based upon sharp changes in blood pressure or heart rate (T3). A 66% overlap of the T1 and T2 segments is also required for an event to be classified as AD. After all parameters have been evaluated for a potential event, and the event has been classified as AD, the event is plotted and saved for any later visual confirmation that may be needed. To validate the automated algorithm, the same data was analyzed using both the semi-automated and fully automated techniques. The results are shown below in Figure 1C. The two methods yield similar results, with the fully automated technique typically detecting more events than the semi-automated technique. This discrepancy is due to both the increased sensitivity of the fully automated technique, as well as an increased number of false positive detections.

Colorectal distension and pinch to intentionally elicit AD: Colorectal distension and cutaneous pinch were used to elicit spinal autonomic reflexes as described previously [9]. The tips of Hartman hemostats were shielded with polyethylene tubing and then used to pinch the flank below the level of SCI just rostral to the hip joint. To ensure consistent pinch intensity and duration, the hemostat was closed to the first click in every trial for 30 s. Colorectal distension was accomplished using a 4-French, 60 mm balloon-tipped catheter (Swan-Ganz monitoring catheter model 116F4; Edwards Life Sciences). The catheter was inserted into the anus, positioning the balloon ~1.5 cm from the anal opening and then securing the catheter to the tail with surgical tape. After securing the catheter, animals were left alone to acclimate for at least 20 min. To elicit AD, the balloon was inflated with 0.3 ml of air for 1 min. Distention was maintained for 1 min and repeat stimulation occurred after a 30 min rest. Peak changes in BP and corresponding HR were obtained and then compared with baseline values.

2) Significant results or key outcomes

Automated detection of AD events was established use a MATLAB program and validated by comparing with the semi-automated techniques (Figure 1). With this automated detection method we can easily analyze continuous cardiovascular data (e.g., MBP and MHR) over 35 days for 16 mice (limited by the number of transmitter receiver) within hours (as compared to

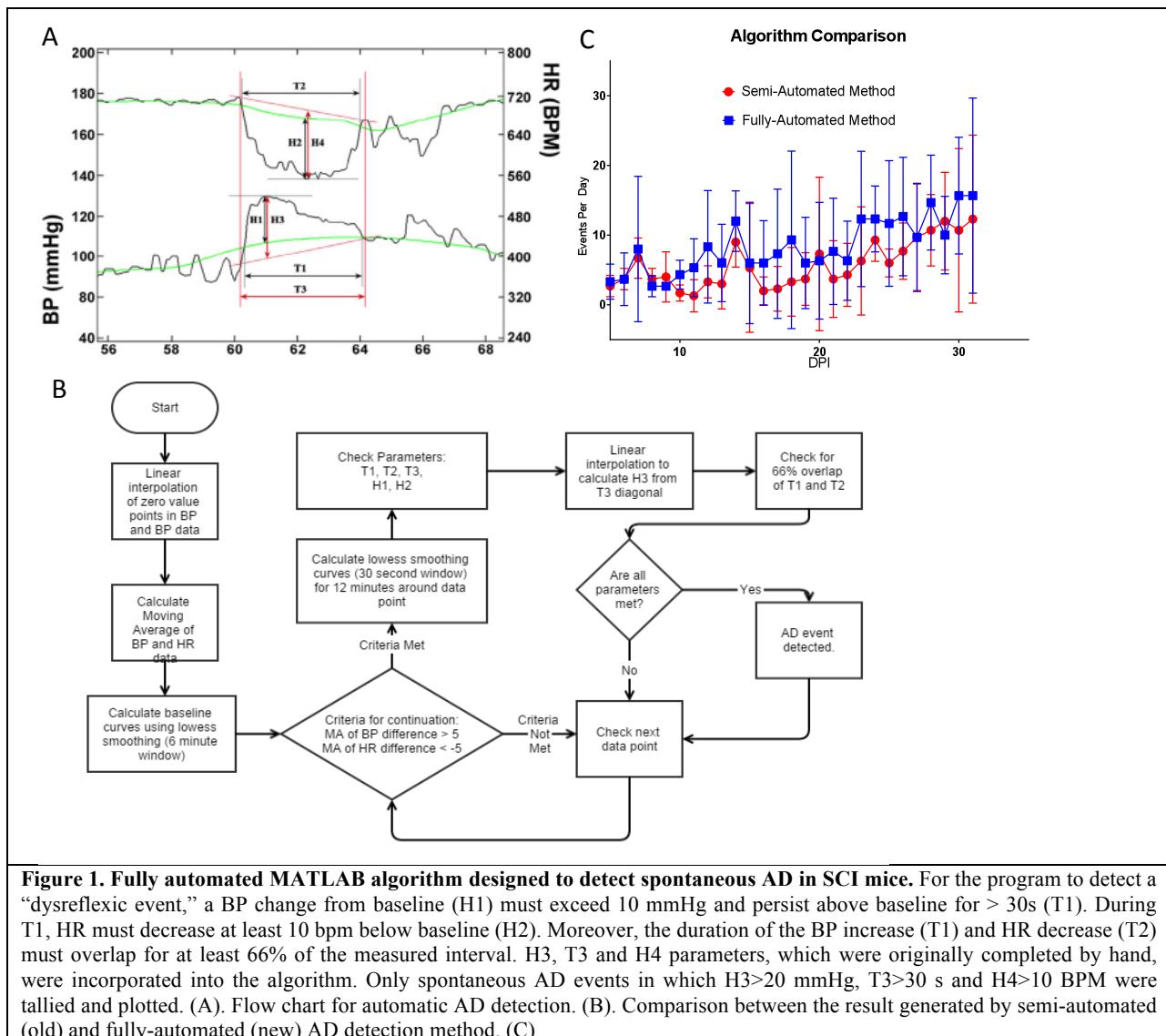
weeks or months with previous method). Overall, the automated program works similarly to the previous semi-automated method, but incorporates the manual validation steps into the computer algorithm to improve objectivity and efficiency.

Both a2d1 TG and WT littermates, which are on a 129 genetic background, did not develop late phase autonomic dysreflexia as described previously in WT C57BL/6J (B6) mice. Also, when compared with WT littermate control mice, the number of spontaneously occurring AD events increases significantly in a2d1 TG mice during the first week post-injury after T3 SCI (Figure 2). These data indicate genetically-encoded differences in mechanisms controlling autonomic dysfunction after SCI.

To determine whether it is possible to intentionally elicit AD in 129 mice, skin pinch or colorectal distension were used to elicit somatic or visceral-sympathetic reflexes below the level of injury. Such stimuli are potent inducers of autonomic dysreflexia after high-level SCI. Surprisingly, neither stimulus elicited AD in most (~80%) 129 mice (Figure 3).

Previous data from our lab indicate that high-level SCI (e.g., T3 injuries) causes immune suppression, an effect that is linked to the development of aberrant autonomic reflexes in the isolated spinal cord below the level of injury [9]. Although we were unable to elicit AD in 129 mice, we next determined whether immune suppression develops in these mice after T3 SCI. The spleens of both a2d1 TG, and WT mice were isolated at 38 days post-injury and spleen weights were normalized to body weight. The data are consistent with our previous observations in C57BL/6 SCI mice, i.e., there is significant splenic atrophy after T3 SCI in both a2d1 TG and WT 129 animals [9] (Figure 4). From these data we conclude that post-injury plasticity in the autonomic circuitry responsible for AD and immune suppression can be mutually exclusive.

Both TSP4 KO and WT littermates, which are on a B6/129P2 genetic background, developed significant amount of AD even at first week post injury. For the TSP4 KO mice the early phase of AD lasted up to 20 days post injury and the late phase started from 27 days post injury. For WT littermate the early phase last until the end of experiment (33dpi) (Figure 5). We speculated that the second phase of AD developed beyond 35dpi, which can't be detected by our current experiment set up (the battery of PA-C10 telemetry transmitters only last for 30-35 days). Interestingly, there is strong correlation between the occurring of AD and the bladder care. The number of AD 1hr before bladder care is significantly higher than after bladder care (Figure 6) can be explained that extended bladder will trigger more AD events even at early phase of AD developing. We speculated that even at the early phase (1-2 week post injury), aberrant collateral sprouting or exist abnormal sensory circuitry may involved in the development of AD.



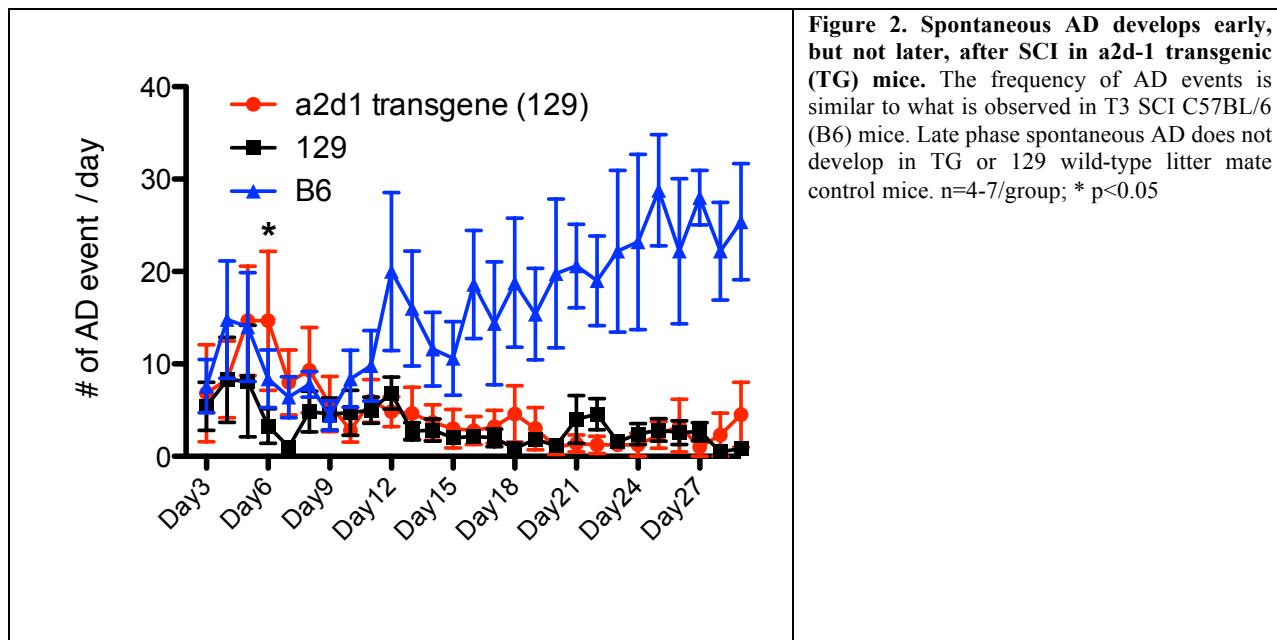


Figure 2. Spontaneous AD develops early, but not later, after SCI in a2d1 transgenic (TG) mice. The frequency of AD events is similar to what is observed in T3 SCI C57BL/6 (B6) mice. Late phase spontaneous AD does not develop in TG or 129 wild-type litter mate control mice. n=4-7/group; *p<0.05

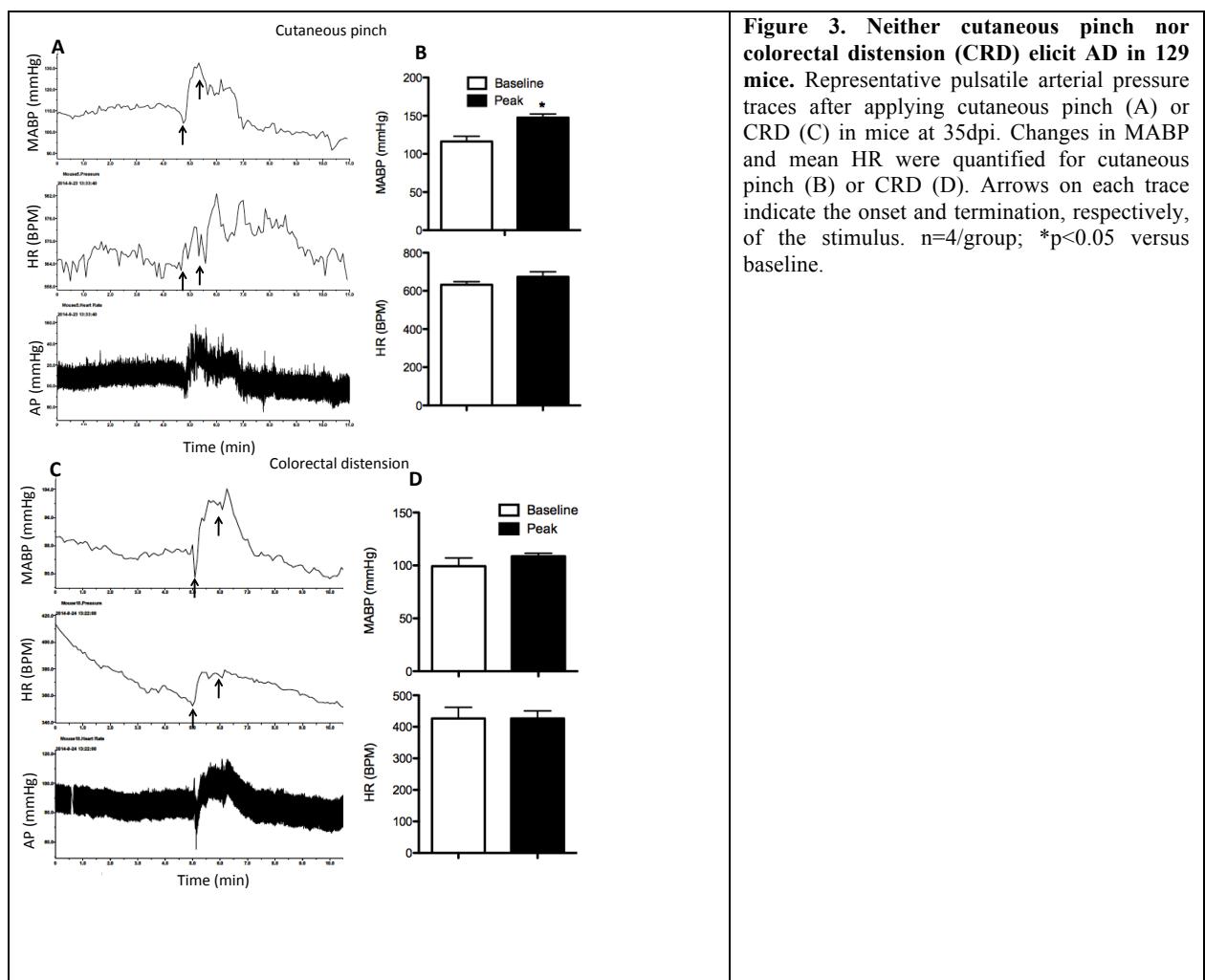
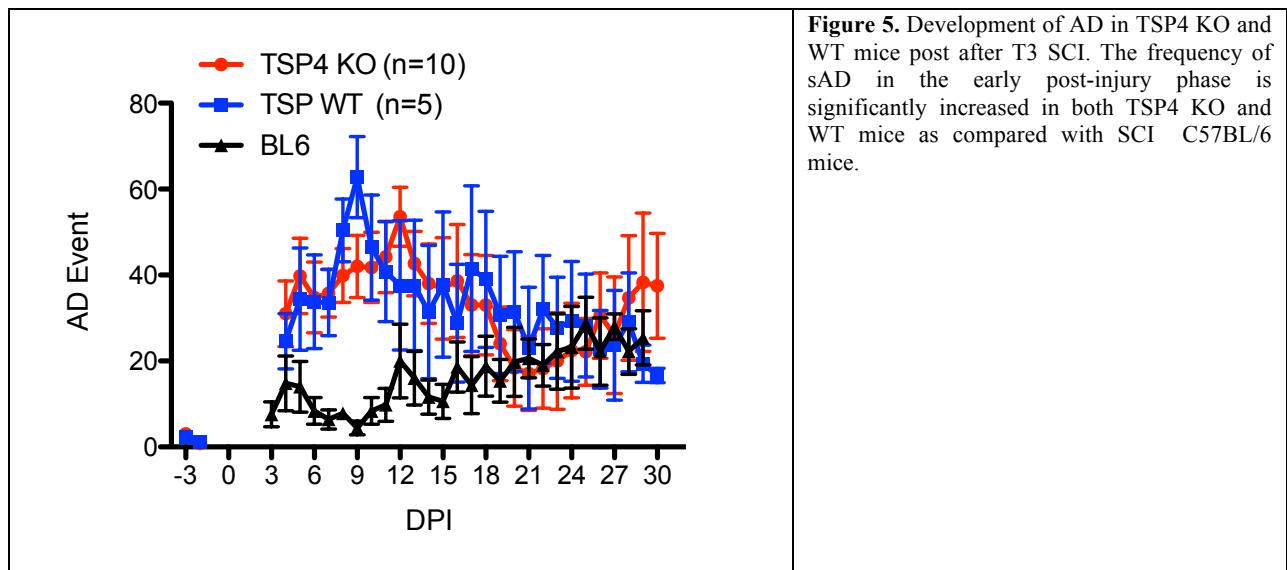
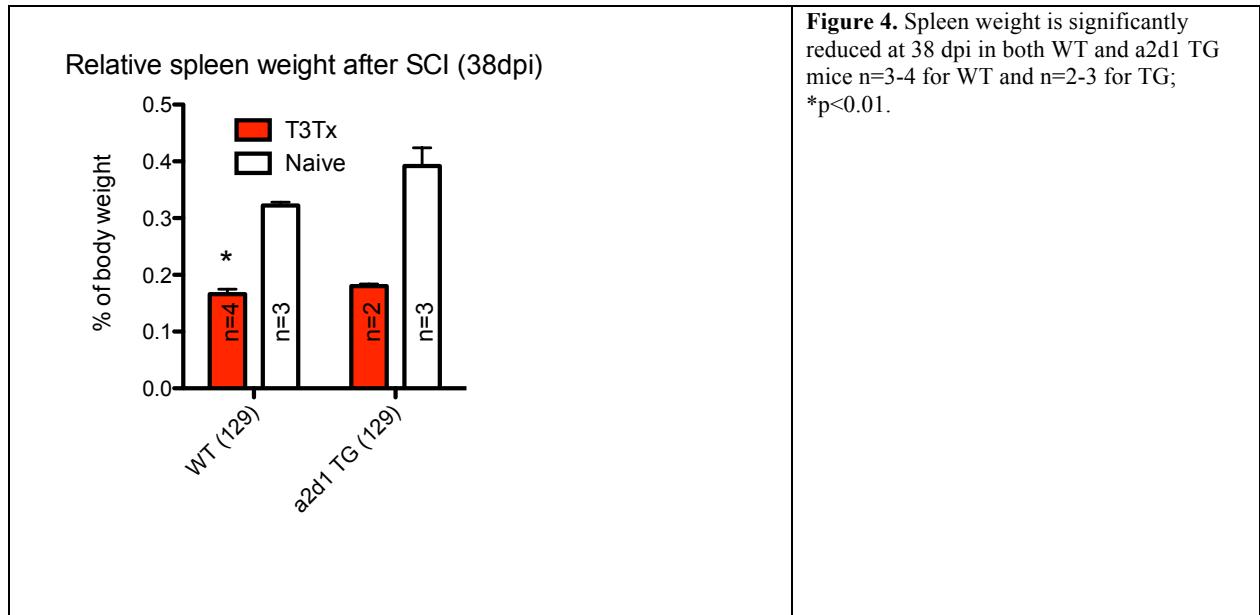
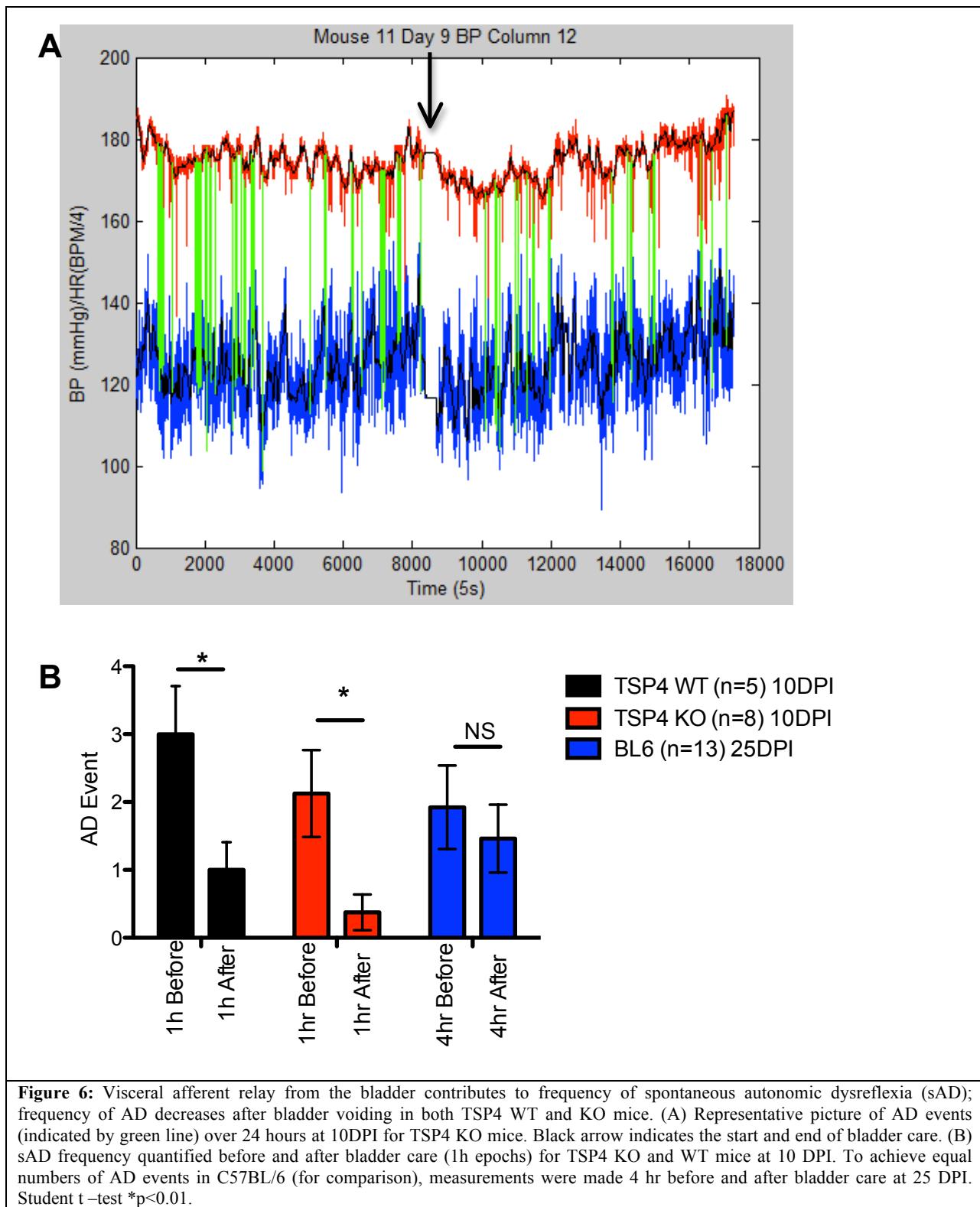


Figure 3. Neither cutaneous pinch nor colorectal distension (CRD) elicit AD in 129 mice. Representative pulsatile arterial pressure traces after applying cutaneous pinch (A) or CRD (C) in mice at 35dpi. Changes in MABP and mean HR were quantified for cutaneous pinch (B) or CRD (D). Arrows on each trace indicate the onset and termination, respectively, of the stimulus. n=4/group; *p<0.05 versus baseline.



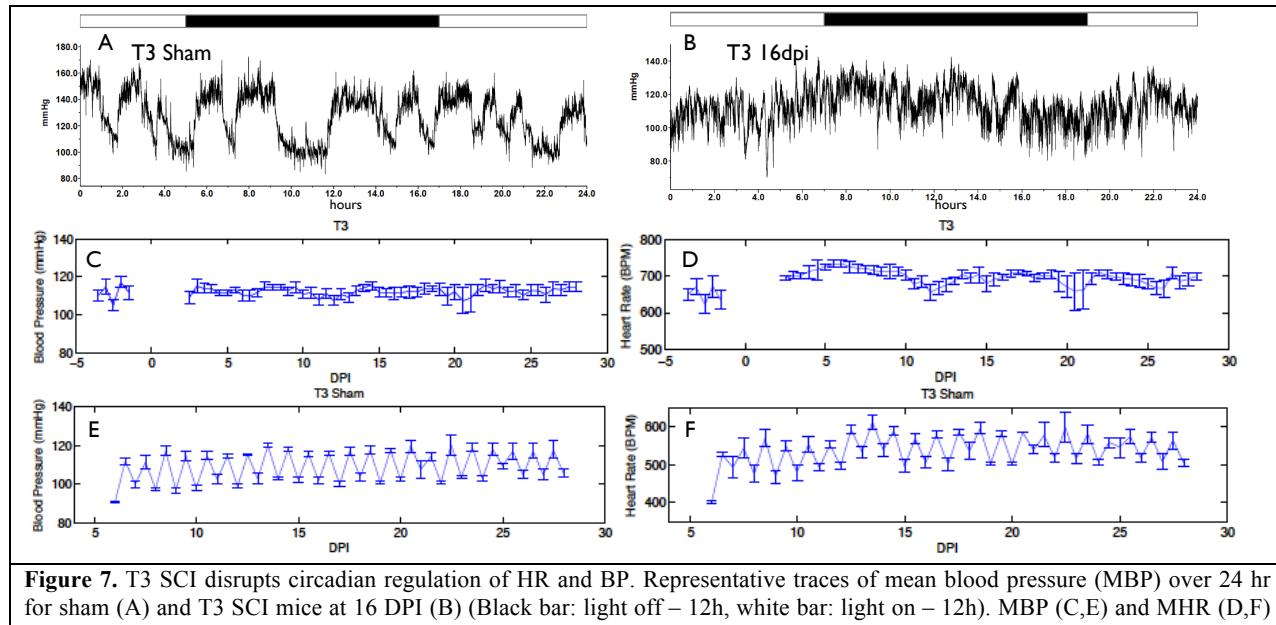


3) Other achievements.

Post-injury disruption of circadian control over cardiovascular function. Using the C57BL/6

mouse model of T3 SCI with telemetry to measure HR and BP, we have found that circadian control over cardiovascular function is significantly impaired after (Figure 7)

Specifically, bimodal distributions of blood pressure and heart rate are disrupted after T3 SCI. The histograms are generated with corresponding mean heart rate (MHR) and mean blood pressure (MBP) (5s average) over 12h intervals corresponding with day and night. For sham animals, the distribution of MBP and MHR show two distinct modes - a resting mode and active mode [11, 12]. T3 SCI disrupts this bimodal distribution of BP and HR and shifts the cardiovascular function to a single mode. Published data indicate that in normal mice, the greatest source of MBP and MHR variation is due to behavioral activity. Although mice are nocturnally active animals, they are also moderately active during the “lights-on” period. During this time, HR and BP values increase such that they approach the values found during the “lights-off” period. Consequently, in normal mice, MBP for both nighttime and daytime were clustered around two modes – resting and active. MHR was significantly higher in mice with T3 injury than sham injured mice and the HR variation over daytime and nighttime was significantly reduced in mice after T3 SCI (Figure 8).



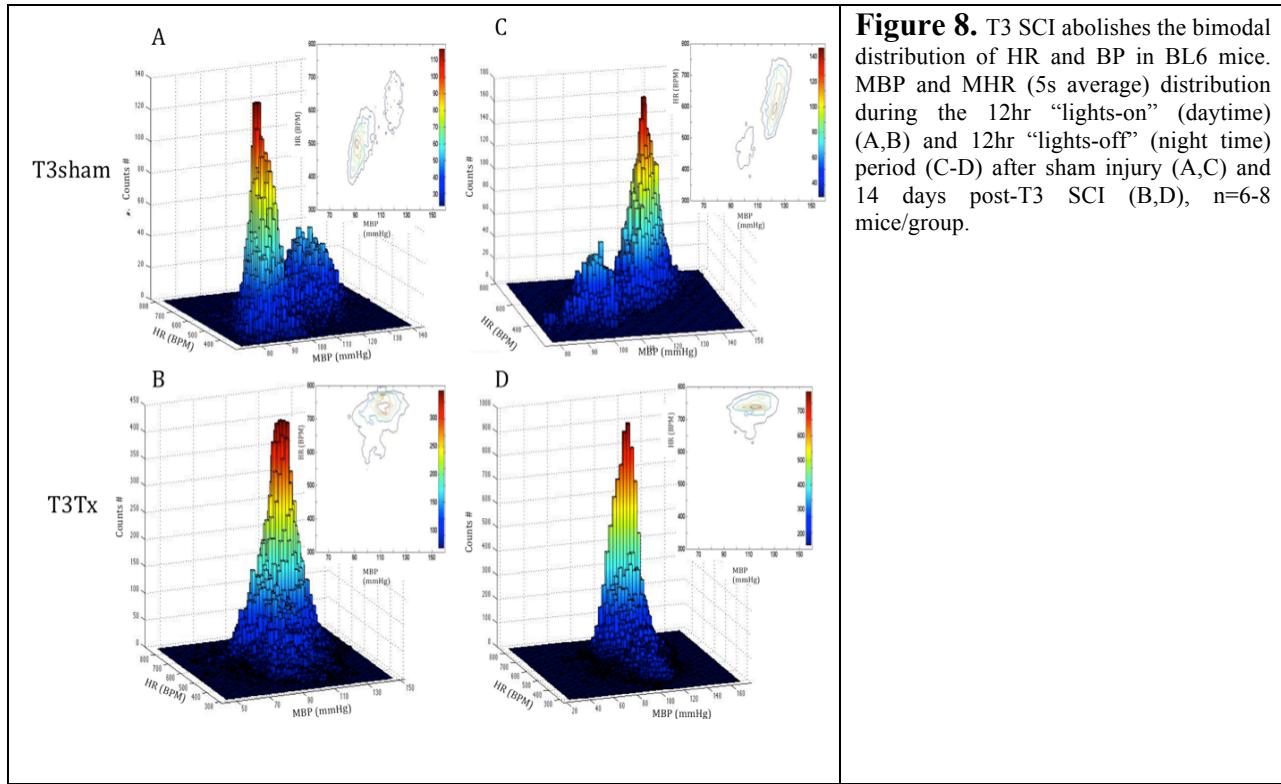
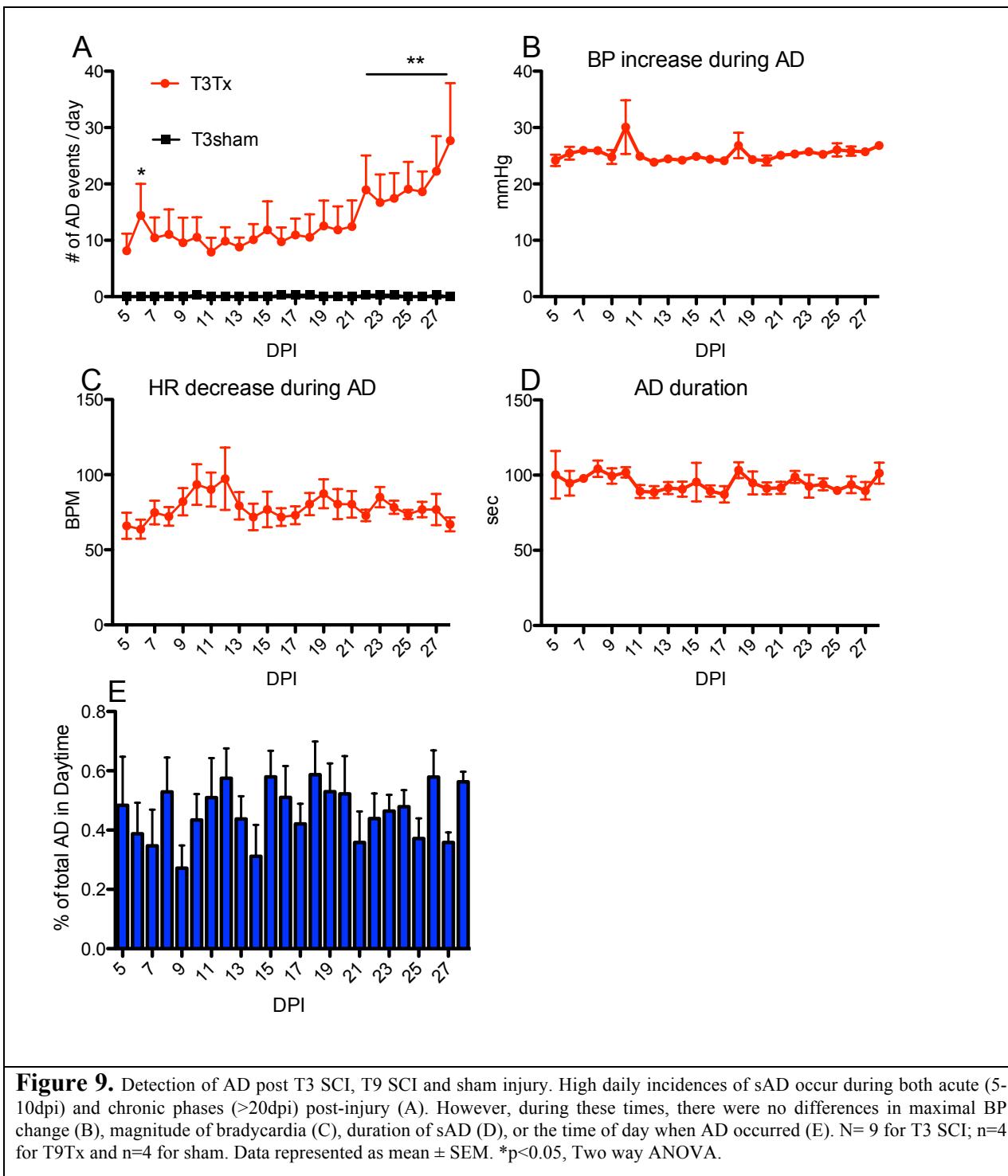


Figure 8. T3 SCI abolishes the bimodal distribution of HR and BP in BL6 mice. MBP and MHR (5s average) distribution during the 12hr “lights-on” (daytime) (A,B) and 12hr “lights-off” (night time) period (C-D) after sham injury (A,C) and 14 days post-T3 SCI (B,D), n=6-8 mice/group.

Using the novel AD detection algorithm described above (see Fig. 1), we were able to extract additional important data related to the onset and progression of spontaneous AD including the maximal increase of BP during periods of AD, maximal drop of HR during those same periods of AD, duration of each dysreflexic event and the time during the day/night when AD develops. There is slight increase number of AD at first week post T3 SCI and from 24 to 28 DPI comparing with sham (Fig. 9A) which is consistent with our previous report [13]. There was no significant difference across time for the severity of AD (drop of BP), the degree of AD-induced bradycardia, the duration of AD events, or whether AD occurred predominantly during the light or dark period (Fig. 9 B, C, D, E).



Developing and characterizing an incomplete model of severe SCI: A new model of SCI was developed and tested. Previously, we used a complete T3 transection injury in order to ensure consistent development of autonomic dysreflexia. Animals received laminectomies after which the periosteum and dura mater were opened with forceps and microscissors. Iridectomy scissors, together with gentle aspiration, were used to sever the spinal cord until a clear separation was

visible between the rostral and caudal stumps of the spinal cord. This surgical procedure is time consuming and requires significant post-surgical animal care. In the past year, we tested a simplified more efficient method of T3 SCI. #4 Dumont tweezers with blunt tip were used to generate a complete spinal crush injury. The forcep tips were inserted bilaterally between the T3 and T4 vertebrae until both tips touch the inner/ventral aspects of the spinal column (ventral-most limit of insertion). After stabilizing the forceps in position, forceps are compressed completely and held in place for 3 seconds then the spinal cord is released. Compared with the previous cut-and-aspiration method, the complete crush injury generates a lesion of similar severity but without compromising the dura or laminae. This reduces surgical time and animal stress. Importantly, the new T3 crush model generates lesions that more closely mimic the human condition while still producing consistent spontaneous AD (see Fig. 10).

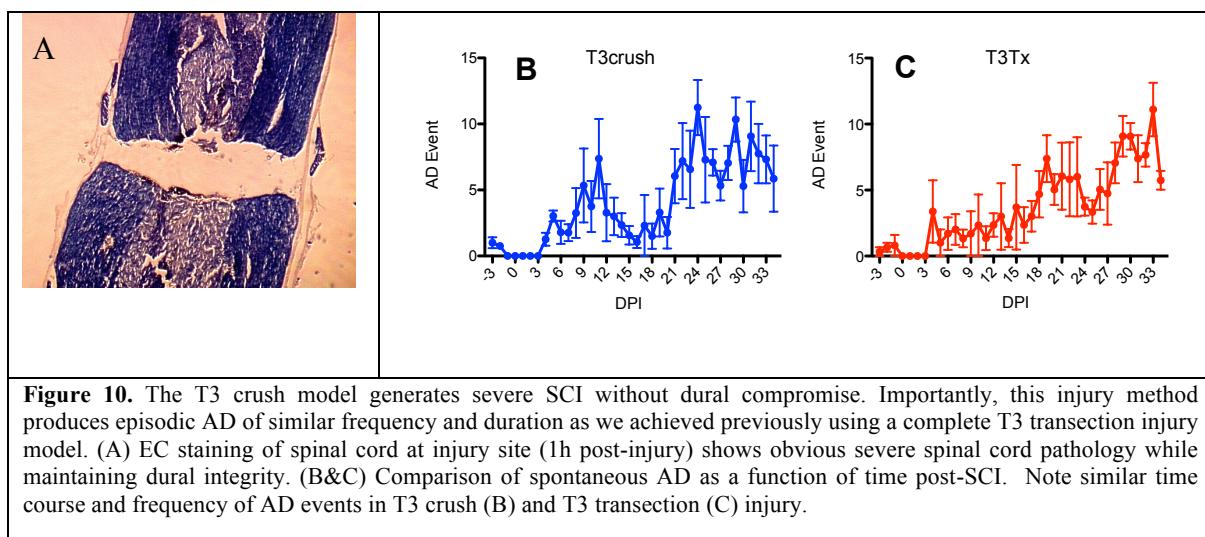


Figure 10. The T3 crush model generates severe SCI without dural compromise. Importantly, this injury method produces episodic AD of similar frequency and duration as we achieved previously using a complete T3 transection injury model. (A) EC staining of spinal cord at injury site (1h post-injury) shows obvious severe spinal cord pathology while maintaining dural integrity. (B&C) Comparison of spontaneous AD as a function of time post-SCI. Note similar time course and frequency of AD events in T3 crush (B) and T3 transection (C) injury.

Goal 2: Comparing AD events in GBP vs control group after T3 SCI. (2% completion)

1) Major activities and Specific objectives:

Pharmaceutical grade (99% certified purity) gabapentin was purchased from Sigma-Aldrich. We then tested the bio-distribution of gabapentin in spinal cord, comparing peripheral delivery by mini-pump or via daily subcutaneous (s.c.) injection.

2) Significant results or key outcomes

Mass spectroscopy was used to measure intraspinal GBP concentrations after daily delivery via s.c. injection (200mg/kg/day) or constant infusion via s.c. mini-pump (0.12ug/6ul/day - maximum possible dose via mini-pump) (Figure 11). The data indicate that higher GBP concentrations are achieved using daily s.c. injections. Even 32h after injecting GBP, the concentration in spinal cord is twice as high as can be achieved via mini-pump over the course of 1 week. The t_{1/2} for GBP in spinal cord was also calculated to be t_{1/2}=9.7h. From these data, to achieve the desired GBP concentration in spinal cord, GBP will be injected s.c. 3x/day for each mouse.

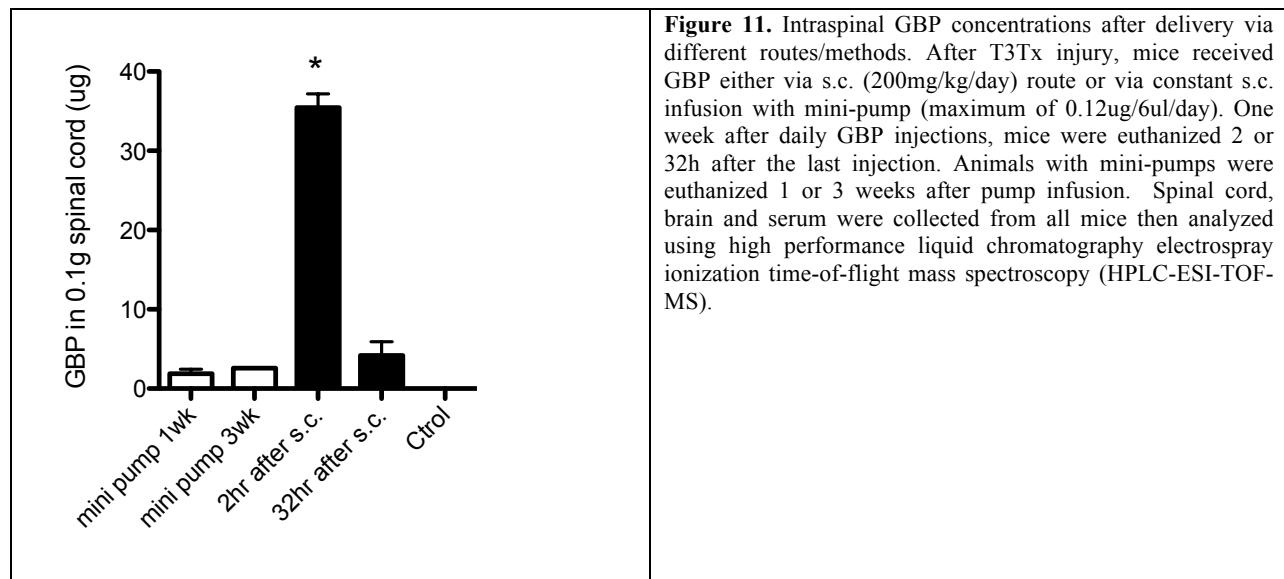


Figure 11. Intraspinal GBP concentrations after delivery via different routes/methods. After T3Tx injury, mice received GBP either via s.c. (200mg/kg/day) route or via constant s.c. infusion with mini-pump (maximum of 0.12ug/6ul/day). One week after daily GBP injections, mice were euthanized 2 or 32h after the last injection. Animals with mini-pumps were euthanized 1 or 3 weeks after pump infusion. Spinal cord, brain and serum were collected from all mice then analyzed using high performance liquid chromatography electrospray ionization time-of-flight mass spectroscopy (HPLC-ESI-TOF-MS).

3) Other achievements.

Nothing to report.

Goal 3: Complete immunohistochemistry analysis for synaptogenesis of all animals. (15% completion)

1) Major activities and Specific objectives:

BL6 mice (sham vs T3 SCI n=6 each group), a2d1TG (n=5) mice and TSP4KO (n=9) mice as well as their WT littermates (n=7 and n=5) were perfused intracardially with TBS (25mM Tris-base, 135 mM NaCl, 3mM KCl, pH7.6) supplemented with 7.5 uM heparin followed with 4% PFA in TBS at 35 DPI. The brains and spinal cord were removed and fixed with 4% PFA in TBS at 4C overnight and shipped to Dr Eroglu (Duke University) for further analysis for the synapse genesis. Using tissues from these mice, synapse analyses within the M1 cortices of the TSP4KO and WT mice (both layer 1 and layer 5) is in progress. Spinal cord analysis is pending.

2) Significant results or key outcomes

Nothing to report.

3) Other achievements.

Nothing to report.

What opportunities for training and professional development has the project provided?

Nothing to Report

How were the results disseminated to communities of interest?

Nothing to Report

What do you plan to do during the next reporting period to accomplish the goals?

For the next reporting period, we plan to repeat the a2d1 TG vs WT and TSP4KO vs WT experiments to confirm the data. Also, we will start experiments in Aim 2 (Goal 2). Specifically, we will test whether prophylactic gabapentin (GBP) injection s.c. tid (three times per day) can reduce post-injury synaptogenesis and the frequency and/or severity of AD.

IMPACT

What was the impact on the development of the principal disciplines of the project?

Our preliminary study indicate that the late phase of AD that develops ~2 weeks after T3 SCI in C57BL/6 mice is eliminated or at least reduced in 129 mice and might be delayed in B6/129p2 mice. The average AD /day/mice during the first 35 days post injury is significantly different between mice with different genetic background (Figure 12). From these data we conclude that susceptibility to the development of AD after SCI may have a previously unrecognized genetic component [14, 15].

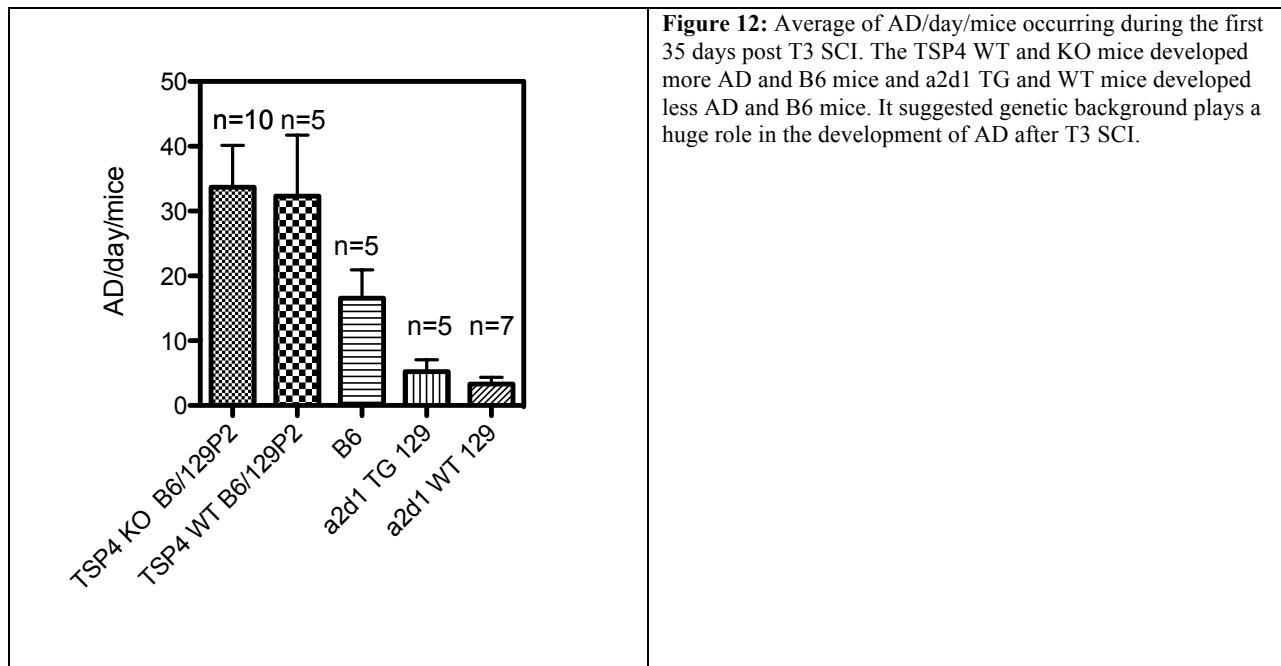


Figure 12: Average of AD/day/mice occurring during the first 35 days post T3 SCI. The TSP4 WT and KO mice developed more AD and B6 mice and a2d1 TG and WT mice developed less AD and B6 mice. It suggested genetic background plays a huge role in the development of AD after T3 SCI.

What was the impact on other disciplines?

Nothing to report

What was the impact on technology transfer?

Nothing to report

What was the impact on society beyond science and technology?

Nothing to report

CHANGES/PROBLEMS

Changes in approach and reasons for change

We simplified and improved our model of spinal cord injury (see Fig. 10 above and accompanying text).

Actual or anticipated problems or delays and actions or plans to resolve them

We do not have a2d1KO mice. This part of the program is unlikely to be developed in the next year.

Changes that has a significant impact on expenditures

Nothing to report

Significant changes in use or care of human subjects, vertebrate animals biohazards, and/or select agents

Nothing to report

Significant changes in use or care of human subjects

NA

Significant changes in use or care of vertebrate animals

NA

Significant changes in use of biohazards and/ or select agents.

NA

PRODUCTS

Publications, conference papers, and presentations

A manuscript describing the cardiovascular dysfunction and changes in circadian regulation in C57BL/6 mice after SCI has been written and will be submitted within the next 3-4 weeks.

Website or other Internet site

Nothing to report

Technologies or techniques

An autonomic AD detection algorithm was developed in this study. The computer code has been made freely available to colleagues.

Invention, patent applications, and/or licenses

Nothing to report

Other products

Nothing to report

PARTICIPANTS & OTHER COLLABORATING ORGANIZATIONS

What individuals have worked on the project?

Name:	Yan Wang
Project Role:	Postdoc Researcher
Researcher Identifier	200143183
Nearest person month worked	12
Contribution to project:	Performing experiment, data analysis
Funding support:	
Name:	Zhen Guan
Project Role:	Senior Research Associate
Researcher Identifier	
Nearest person month worked	12
Contribution to project:	Performing experiment
Funding support:	
Name:	J. Hayes Davis
Project Role:	Undergraduate Researcher
Researcher Identifier	2003187666
Nearest person month worked	10
Contribution to project:	data analysis
Funding support:	
Name:	Phillip Popovich
Project Role:	Professor
Researcher Identifier	
Nearest person month worked	12
Contribution to project:	Design the experiment, instruction.
Funding support:	

Has there been a change in the active other support of the PI or key personnel since the last reporting period?

Nothing to report

What other organizations were involved as partners?

Organization Name: Duke University Medical Center, Department of Cell Biology

Location: 334 Nanaline Duke Building, Durham, NC 27710

Collaboration: Dr. Cagla Eroglu, Ph.D.

SPECIAL REPORTING REQUIREMENTS

Collaborative awards:

For collaborative awards, independent reports are required from BOTH the Initiating PI and the Collaborating/Partnering PI. A duplicative report is acceptable; however, tasks shall be clearly marked with the responsible PI and research site. A report shall be submitted to <https://ers.amedd.army.mil> for each unique award

QUAD CHARTS:

If applicable, the Quad Chart (available on <https://www.usamraa.army.mil>) should be updated and submitted with attachments.

Attached in the end.

APPENDICES

Attach all appendices that contain information that supplements, clarifies or supports the text. Examples include original copies of journal articles, reprints of manuscripts and abstracts, a curriculum vitae, patent applications, study questionnaires, and surveys, etc

1. Weaver, LC, Fleming, JC, Mathias, CJ, and Krassioukov, AV. Disordered cardiovascular control after spinal cord injury. *Handb Clin Neurol* 109: 213-233.
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4. Hou, S, Duale, H, Cameron, AA, Abshire, SM, Lytle, TS, and Rabchevsky, AG (2008). Plasticity of lumbosacral propriospinal neurons is associated with the development of autonomic dysreflexia after thoracic spinal cord transection. *J Comp Neurol* 509: 382-399.
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7. Eroglu, C, et al. (2009). Gabapentin receptor alpha2delta-1 is a neuronal thrombospondin receptor responsible for excitatory CNS synaptogenesis. *Cell* 139: 380-392.
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13. Zhang, Y, et al. Autonomic dysreflexia causes chronic immune suppression after spinal cord injury. *J Neurosci* 33: 12970-12981.
14. van Bogaert, MJ, Groenink, L, Oosting, RS, Westphal, KG, van der Gugten, J, and Olivier, B (2006). Mouse strain differences in autonomic responses to stress. *Genes Brain Behav* 5: 139-149.
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